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Gerald H. Kiel, Esq.
REED SMITH LLP
599 Lexington Avenue
New York, NY 10022-7650

EXAMINER	
FORD, ALLISON M	

ART UNIT	PAPER NUMBER
1651	

DATE MAILED: 11/18/2004

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/734,903	SCHAFFER ET AL.	
	Examiner	Art Unit	
	Allison M Ford	1651	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☐ Responsive to communication(s) filed on _____.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) 1-6 is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) 1-6 is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☒ All b)☐ Some * c)☐ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

Status of Application

Claims 1-6 are pending in the current application.

Priority

Acknowledgement is made of applicant's claim for priority to German national application 102 58989.5, filed 12/13/02; however no translation has been provided.

Claim Rejections - 35 USC § 112

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1-6 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a test preparation for fluorescence microscopes, does not reasonably provide enablement for all optical microscopes.

Applicant's claim 1 is directed to a test preparation for optical microscopes, comprising an object carrier and a cell bond fixed under treatment by a compound which enables a freely selectable fluorescence excitation in a wavelength region with a breadth of the order of 100 nm or greater. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims. Optical microscopes include all microscopes that utilize light to view the specimen, including the common light microscopes, such as bright field and dark field microscopes, inverted light microscopes, phase contrast microscopes, and fluorescence microscopes. The test preparation in the current application uses a compound to produce fluorescence emissions; because the emissions are fluorescent, they can only be properly viewed through a fluorescent microscope because of its unique filters and mirror systems. Therefore, the test preparation cannot be used with any optical microscope;

it can only be used successfully with a fluorescence microscope. Claims 2-6 have the limitations of claim 1, and thus are rejected on the same basis.

Claims 1-6 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

Applicant's claim 1 is directed to a test preparation for optical microscopes, comprising an object carrier and a cell bond fixed under treatment by a compound which enables a freely selectable fluorescence excitation in a wavelength region with a breadth of the order of 100 nm or greater. In the disclosure applicant describes the use of glutardialdehyde for inducing fluorescence in cells. Glutardialdehyde-induced fluorescence is taught in the art (See, e.g. Collins et al and Frank et al); however, in order to claim a genus, such as any compound that would induce fluorescence, a representative number of species must be adequately described and enabled by the specification, see MPEP § 2163. The teaching of use of glutardialdehyde as the single, non-exemplified fluorescence-inducing compound does provide sufficient description of the genus, or constitute a representative number of species, both of which are required to claim all fluorescence-inducing compounds. Additionally, there is no disclosure of relevant, identifying characteristics, such as structure or other physical or chemical properties, or functional characteristics, beyond disclosure of the generic action (fluorescence inducing), sufficient to show the applicant was in possession of the claimed genus. *See Eli Lilly,* 119F. 3d. at 1568, 43 USPQ2d at 1406.

Claims 1-6 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for induced fluorescence by glutardialdehyde, does not reasonably provide enablement for induced fluorescence by any compound. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims. Glutardialdehyde induced fluorescence is known in the art (See, e.g. Collins et al and Frank et al);

however applicant is not enabled to claim any compound, as most compounds do not produce fluorescence or fix cells. For example paramagnetic species such as O_2 and iodide reduce fluorescence (See Molecular Probes), and have no fixing capabilities. Therefore applicant cannot claim any compound for use in preparing the test preparation for microscopes as claimed in claims 1-3. Claims 4-6 have the limitations of claim 1, and thus are rejected on the same basis.

Claims 1-6 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for exhibiting fluorescence excitation at 540 nm does not reasonably provide enablement for exhibiting fluorescence excitation at any wavelength of 100 nm or greater. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims.

Because the specification is directed solely to the use of glutardialdehyde to treat cells, and claims 1-3 and 5-6 are enabled only for glutardialdehyde the following scope of enablement rejection is made with the interpretation that glutardialdehyde is the said compound.

In the disclosure applicant teaches using glutardialdehyde to simultaneously fix and "stain" cells to cause them to fluoresce; they claim the fluorescence due to the glutardialdehyde is capable of being excited by wavelengths 100 nm or greater. However, their complete lack of experimental evidence and working examples does not show that they achieved excitation at any wavelength; in fact, they failed to teach, even through incorporation by reference, why or how the glutardialdehyde induces fluorescence.

Glutardialdehyde induced fluorescence is known and taught in the art, for example, Collins et al teach the stabilizing cross-linking of glutardialdehyde produces the fluorophores that fluoresce (See Pg. 411). Collins performs experiments on crayfish rhaboms, treating the cells with glutardialdehyde and measuring the resulting fluorescence (See Pg. 411); however, Collins reports fluorescence was maximally excited at a wavelength of 540 nm, but fluorescence was achieved over the range of 420-560 nm (See Fig. 1A). Also, Frank et al performs experiments on platelets, treating the cells with glutardialdehyde, and achieving fluorescence in the range of 450-490 nm (See Pg. 376, col. 1).

Therefore, while applicant may claim glutardialdehyde is capable of fluorescence excitation in the range of 420-560 nm, with reference to Collins et al, they are not enabled to for an excitation range of 100 nm or greater, or even 350-700 nm (Claim 3), or even 450-650 nm (Claim 2) without providing evidence that they have successfully excited the fluorophores produced by glutardialdehyde at any wavelength 100 nm or greater, or spanning 350-700 nm, or even 450-650 nm. Claims 4-6 have the limitations of claim 1, and thus are rejected on the same basis.

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 1-6 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Applicant's claim 1 is directed to a test preparation for optical microscopes, comprising an object carrier and a cell bond fixed under treatment by a compound which enables a freely selectable fluorescence excitation in a wavelength region with a breadth of the order of 100 nm or greater. Several terms in this claim are unclear, and thus make the claim confusing and indefinite. First, it is not clear what a "cell bond" consists of, as there does not appear to be any set meaning in the art. The term "cell bond" most closely refers to cell-adhesion molecules, cell-cell junctions, or other specific binding structures between cells; however it appears applicant intended to refer to a cell or a section of tissue. Therefore it is unclear what is being fixed onto the object carrier, the term is being interpreted to mean a cell or a section of tissue. Second, the phrase, "under treatment by a compound" would more appropriately be stated, "fixed by a compound," or "fixed with a compound." Third, it is not clear what is intended by "a freely selectable fluorescence excitation in a wavelength region with a breadth of the order of 100 nm or greater." If applicant intends to claim the fixative compound permits fluorescence excitation in a broad range of wavelengths, including 100 nm and greater, it

needs to be clarified to read as such. Claims 2-6 have the limitations of claim 1 and are rejected on the same basis.

Applicant's claims 2 and 3 are directed to the test preparation for microscopes of claim 1, wherein the cell bond is fixed under treatment by a compound which enables a freely selectable fluorescence excitation in a wavelength range from 450-650 nm (claim 2) or 350-700 nm (claim 3). The term "cell bond" is unclear for the same reason as stated above. Additionally, the phrase, "under treatment by a compound which enables a freely selectable fluorescence excitation in a wavelength range of..." again would be clearer if written "fixed with a compound that permits fluorescence excitation in a wavelength range from ..."

Applicant's claim 4 is directed to the test preparation for microscopes according to claim 1, wherein the cell bond is fixated using glutardialdehyde. Again, the term "cell bond" is unclear for the same reason as stated above. Also, it is unclear if the glutardialdehyde is the compound which the cell bond (being interpreted to mean cell or tissue) is the compound the cell by is "fixed under treatment by," or if it is an additional component.

Applicant's claim 6 is directed to the test preparation for microscopes according to claim 1, wherein the cell bond has a dense structure over the entire visual field of the microscope. The term "cell bond" is unclear for the same reasons as stated above. Furthermore, it is not clear what is meant by the cell bond (being interpreted to mean cell or tissue) having a dense structure over the visual field of the microscope. It is unclear if the structure is that of the cell or tissue, or of an additional dense structure added to the object carrier. If the structure is in reference to the cell or tissue it is unclear how the density of a cell is changed so that it may be made more dense; similarly, a tissue section can be thick or thin, changing the volume of the sample, but this would not affect the density, as D= m/V. For purposes of examination the claim is being interpreted to mean the cells densely cover the entire visual field of the microscope.

Claim Rejections - 35 USC § 102

(b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

Claims 1-5 are rejected under 35 U.S.C. 102(b) as being anticipated by Frank et al (*J. Biomed Mater. Res*, 2000), in light of DakoCytomation.

Applicant's claim 1 is directed to a test preparation for microscopes comprising an object carrier and a biological cell bond arranged on the object carrier, wherein the cell bond is fixed with a compound which enables a freely selectable fluorescence excitation in a broad range of wavelengths, including 100 nm and greater. The cell bond is being interpreted to mean a cell or tissue, comprised of cells. Claim 2 requires the fluorescence excitation range to be from 450 to 650 nm. Claim 3 requires the fluorescence excitation range to be from 350 to 700 nm. Claim 4 requires the cell to be fixed with glutardialdehyde. Claim 5 requires an anti-fading reagent to be added to the compound.

Frank et al teach glutardialdehyde induced fluorescent thrombocytes on a slide for visualization with computer-aided fluorescence microscopy; fluorescing cells (which applicant calls a cell bond) on an object carrier read on what applicant calls a test preparation for a microscope. (See Pg. 375, col. 1). Frank et al use polymer discs as object carriers. The test preparations were prepared by fixing platelets onto polymer discs (which applicant calls object carriers) using 1.5% glutardialdehyde solution in PBS, the glutardialdehyde both fixes the cells to the discs and induces fluorescence in the cells (See Pg. 375, col. 2). After incubation the discs were removed and placed on microscope slides, one drop of fluorescence mounting media was added and the platelets were detected with a fluorescence microscope at excitation levels of 450-490 nm (See Pg 376, col. 1) (Claims 1-4). The fluorescence mounting media was obtained from DAKO (DakoCytomations), the fluorescence mounting media is an anti-fading agent (See DakoCytomation Product Sheet) (Claim 5). The emission was detected at 515 nm (See Pg. 376, col. 2). Therefore the reference anticipates the claimed subject matter.

Claims 1-4 are rejected under 35 U.S.C. 102(b) as being anticipated by Collins et al (*J Histochem Cytochem*, 1981).

Applicant's claim 1 is directed to a test preparation for microscopes comprising an object carrier and a biological cell bond arranged on the object carrier, wherein the cell bond is fixed with a compound which

enables a freely selectable fluorescence excitation in a broad range of wavelengths, including 100 nm and greater. The cell bond is being interpreted to mean a cell or tissue, comprised of cells. Claim 2 requires the fluorescence excitation range to be from 450 to 650 nm. Claim 3 requires the fluorescence excitation range to be from 350 to 700 nm. Claim 4 requires the cell to be fixed with glutardialdehyde.

Collins et al teach tissue and gelatin films with glutardialdehyde-induced fluorescence; fluorescing cells (which applicant calls cell bonds) on an object carrier, such as a slide, reads on what applicant calls a test preparation for a microscope. Collins et al produced the test preparations by fixing crayfish rhaboms with 1.9% glutardialdehyde for 20 min (See Pg. 411, col. 1-2). Collins et al do not explicitly state that the tissue and gelatin films were arranged on an object carrier, but this appears to be an understood detail, as tissue and gelatin films cannot hold their own structure on a microscope stand, and it is common practice in the art to use a microscope slide to observe specimens. The tissue and gelatin films treated with glutardialdehyde were observed on a Zeiss fluorescence microscope, under excitation at 420-560 nm. Optimal excitation was achieved at 540 nm, at which point the emission was 560 nm (See Pg. 411, col. 2- Pg. 413, col. 1 and Fig. 1A) (Claims 1-4).

Claims 1-3 are rejected under 35 U.S.C. 102(b) as being anticipated by Molecular Probes "MitoTracker and MitoFluor Mitochondrion-Selective Probes" Product Data Sheet and Molecular Probes "ER-Tracker Blue-White DPX" Product Data Sheet.

Applicant's claim 1 is directed to a test preparation for microscopes comprising an object carrier and a biological cell bond arranged on the object carrier, wherein the cell bond is fixed with a compound which enables a freely selectable fluorescence excitation in a broad range of wavelengths, including 100 nm and greater. The cell bond is being interpreted to mean a cell or tissue, comprised of cells. Claim 2 requires the fluorescence excitation range to be from 450 to 650 nm. Claim 3 requires the fluorescence excitation range to be from 350 to 700 nm.

Molecular Probes teach preparations of cells stained with fluorescent probes; fluorescing cells (which applicant calls cell bonds) on slides read on what applicant calls test preparations for microscopes. Molecular

Probes sell fluorescent probes that can be used to produce slides with stained cells for visualization of cellular components. For example, the family of MitoTracker Dyes are mitochondrion-selective probes that can be used to produce slides that show the mitochondria. Molecular Probes teach incubating cells in presence of Orange-, Red-, Deep Red-, and Green-Fluorescent MitoTracker Dyes and then fixing the cells in an aldehyde-based fixative (See Pg 1, col. 2- Pg. 2, col. 1). MitoTracker Orange exhibits fluorescent excitation at 554 nm; MitoTracker Red exhibits fluorescent excitation at 579 nm; MitoTracker Deep Red exhibits fluorescent excitation at 644 nm; and MitoTracker Green exhibits fluorescent excitation at 490 nm (See Table 1, Pg. 2) (Claims 1-3). Another example, ER-Tracker Blue-White DPX stains the endoplasmic reticulum of eukaryotic cells. Molecular Probes teaches adherent cells can be grown on a coverslip, which acts as an object carrier; incubated with the probe-containing medium for approximately 30 min (See Pg. 1, col. 2). The cells can then be fixed to the cover slip by application of 3.7% formaldehyde for 10-20 min (See Pg 2, col. 2). Fluorescence emission peaks anywhere in the range from 430-640 nm (See Pg. 1, col. 2) (Claims 1-3). Therefore the reference anticipates the claimed subject matter.

Claim Rejections - 35 USC § 102/103

Claim 6 is rejected under 35 U.S.C. 102(b) as being anticipated by or, in the alternative, under 35 U.S.C. 103(a) as being obvious over Frank et al (*J. Biomed Mater. Res*, 2000).

Applicant's claim 1 is directed to a test preparation for microscopes comprising an object carrier and a biological cell bond arranged on the object carrier, wherein the cell bond is fixed with a compound which enables a freely selectable fluorescence excitation in a broad range of wavelengths, including 100 nm and greater. The cell bond is being interpreted to mean a cell or tissue, comprised of cells. Claim 6 requires the cells to densely cover the entire visual field of the microscope.

Frank et al teach glutardialdehyde induced fluorescent thrombocytes on a slide for visualization with computer-aided fluorescence microscopy; fluorescing cells (which applicant calls cell bonds) on an object carrier read on what applicant calls a test preparation for a microscope. (See Pg. 375, col. 1). Frank et al use

polymer discs as object carriers. The test preps were prepared by fixing platelets onto polymer discs (which applicant calls object carriers) using 1.5% glutardialdehyde solution in PBS, the glutardialdehyde both fixes the cells to the discs and induces fluorescence in the cells (See Pg. 375, col. 2). After incubation the discs were removed and placed on microscope slides, one drop of fluorescence mounting media, an anti-fading agent (See DakoCytomation Product Sheet), was added and the platelets were detected with a fluorescence microscope at excitation levels of 450-490 nm (See Pg 376, col. 1) (Claims 1-5). The emission was detected at 515 nm (See Pg. 376, col. 2) (Claim 1).

Frank et al is silent on the density of the platelets plated on the discs. However, it appears the cell density, as claimed, is the same as in the prior art, and thus Frank et al anticipates the subject matter. However, even if the reference cell density and the claimed cell density are not one and the same and there is, in fact, no anticipation, the reference cell density would, nevertheless, have rendered to one of ordinary skill in the art at the time the invention was made the claimed density of the plated cells an obvious design choice based on availability of cells and optimization. The degree of fluorescence is a result effective variable, dependent on the density of stained cells present in the observed sample. Therefore, the density of the plated cells would be routinely optimized by one of ordinary skill in the art in practicing the invention to obtain the desired fluorescent effect.

Thus, the claimed invention as a whole was at least prima facie obvious, if not anticipated by the references, especially in the absence of evidence to the contrary.

Claim 6 is rejected under 35 U.S.C. 102(b) as being anticipated by or, in the alternative, under 35 U.S.C. 103(a) as being obvious over Collins et al (*J Histochem Cytochem*, 1981).

Applicant's claim 1 is directed to a test preparation for microscopes comprising an object carrier and a biological cell bond arranged on the object carrier, wherein the cell bond is fixed with a compound which enables a freely selectable fluorescence excitation in a broad range of wavelengths, including 100 nm and

greater. The cell bond is being interpreted to mean a cell or tissue, comprised of cells. Claim 6 requires the cells to densely cover the entire visual field of the microscope.

Collins et al teach tissue and gelatin films with glutardialdehyde-induced fluorescence; fluorescing cells (which applicant calls cell bonds) on an object carrier, such as a slide, reads on what applicant calls a test preparation for a microscope. Collins et al produced the test preparations by fixing crayfish rhaboms with 1.9% glutardialdehyde for 20 min (See Pg. 411, col. 1-2). Collins et al do not explicitly state that the tissue and gelatin films were arranged on an object carrier, but this appears to be an understood detail, as tissue and gelatin films cannot hold their own structure on a microscope stand, and it is common practice in the art to use a microscope slide to observe specimens. The tissue and gelatin films treated with glutardialdehyde were observed on a Zeiss fluorescence microscope, under excitation at 420-560 nm. Optimal excitation was achieved at 540 nm, at which point the emission was 560 nm (See Pg. 411, col. 2- Pg. 413, col. 1 and Fig. 1A) (Claim 1).

Collin et al is silent on the density of the tissue or gelatin films. However, it appears the cell density, as claimed, is the same as in the films in the prior art, and thus Collins et al anticipates the subject matter.

However, even if the reference cell density of the tissue films and the claimed cell density are not one and the same and there is, in fact, no anticipation, the reference cell density of the tissue films would, nevertheless, have rendered to one of ordinary skill in the art at the time the invention was made the claimed cell density an obvious design choice based on availability of cells and optimization. The degree of fluorescence is a result effective variable, dependent on the density of stained cells present in the observed sample. Therefore, the number of cells in the tissue films would be routinely optimized by one of ordinary skill in the art in practicing the invention to obtain the desired fluorescent effect.

Thus, the claimed invention as a whole was at least prima facie obvious, if not anticipated by the references, especially in the absence of evidence to the contrary.

Claim 6 is rejected under 35 U.S.C. 102(b) as being anticipated by or, in the alternative, under 35 U.S.C. 103(a) as being obvious over Molecular Probes "MitoTracker and MitoFluor Mitochondrion-Selective Probes" Product Data Sheet and Molecular Probes "ER-Tracker Blue-White DPX" Product Data Sheet.

Applicant's claim 1 is directed to a test preparation for microscopes comprising an object carrier and a biological cell bond arranged on the object carrier, wherein the cell bond is fixed with a compound which enables a freely selectable fluorescence excitation in a broad range of wavelengths, including 100 nm and greater. The cell bond is being interpreted to mean a cell or tissue, comprised of cells. Claim 6 requires the cells to densely cover the entire visual field of the microscope.

Molecular Probes teach preparations of cells stained with fluorescent probes; fluorescing cells (which applicant calls cell bonds) on slides read on what applicant calls test preparations for microscopes. Molecular Probes sell fluorescent probes that can be used to produce slides with stained cells for visualization of cellular components. For example, the family of MitoTracker Dyes are mitochondrion-selective probes that can be used to produce slides that show the mitochondria. Molecular Probes teach incubating cells in presence of Orange-, Red-, Deep Red-, and Green-Fluorescent MitoTracker Dyes and then fixing the cells in an aldehyde-based fixative (See Pg 1, col. 2- Pg. 2, col. 1). MitoTracker Orange exhibits fluorescent excitation at 554 nm; MitoTracker Red exhibits fluorescent excitation at 579 nm; MitoTracker Deep Red exhibits fluorescent excitation at 644 nm; and MitoTracker Green exhibits fluorescent excitation at 490 nm (See Table 1, Pg. 2) (Claim 1). Another example, ER-Tracker Blue-White DPX stains the endoplasmic reticulum of eukaryotic cells. Molecular Probes teaches adherent cells can be grown on a coverslip, which acts as an object carrier; incubated with the probe-containing medium for approximately 30 min (See Pg. 1, col. 2). The cells can then be fixed to the cover slip by application of 3.7% formaldehyde for 10-20 min (See Pg 2, col. 2). Fluorescence emission peaks anywhere in the range from 430-640 nm (See Pg. 1, col. 2) (Claim 1).

Molecular Probes is silent on the density or confluency of the cells to be stained. However, it appears the cell density/confluency, as claimed, is the same as in the prior art, and thus Molecular Probes anticipates the subject matter.

However, even if the reference cell density/confluency and the claimed cell density are not one and the same and there is, in fact, no anticipation, the reference cell density/confluency would, nevertheless, have rendered to one of ordinary skill in the art at the time the invention was made the claimed density/confluency of the plated cells an obvious design choice based on availability of cells and optimization, as the degree of fluorescence is a result effective variables, dependent on the density of stained cells present in the observed sample. Therefore, altering the density of the plated cells would have been an obvious design choice easily manipulated to obtain the desired fluorescent effect.

Thus, the claimed invention as a whole was at least prima facie obvious, if not anticipated by the references, especially in the absence of evidence to the contrary.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

Claims 1-5 are rejected under 35 U.S.C. 103(a) as being unpatentable over Collins et al (*J Histochem Cytochem*, 1981), in view of DakoCytomation.

Applicant's claim 1 is directed to a test preparation for microscopes comprising an object carrier and a biological cell bond arranged on the object carrier, wherein the cell bond is fixed with a compound which enables a freely selectable fluorescence excitation in a broad range of wavelengths, including 100 nm and greater. The cell bond is being interpreted to mean a cell or tissue, comprised of cells. Claim 2 requires the fluorescence excitation range to be from 450 to 650 nm. Claim 3 requires the fluorescence excitation range to be from 350 to 700 nm. Claim 4 requires the cell to be fixed with glutardialdehyde. Claim 5 requires an anti-fading reagent to be added to the compound.

Collins et al teach tissue and gelatin films with glutardialdehyde-induced fluorescence; fluorescing cells (which applicant calls cell bonds) on an object carrier, such as a slide, reads on what applicant calls a test preparation for a microscope. Collins et al produced the test preparations by fixing crayfish rhaboms with 1.9% glutardialdehyde for 20 min (See Pg. 411, col. 1-2). Collins et al do not explicitly state that the tissue and gelatin films were arranged on an object carrier, but this appears to be an understood detail, as tissue and gelatin films cannot hold their own structure on a microscope stand and it is common practice in the art to use a microscope slide to observe specimens. The tissue and gelatin films treated with glutardialdehyde were observed on a Zeiss fluorescence microscope, under excitation at 420-560 nm. Optimal excitation was achieved at 540 nm, at which point the emission was 560 nm (See Pg. 411, col. 2- Pg. 413, col. 1 and Fig. 1A) (Claims 1-4).

Though Collins et al does not teach use of an anti-fading reagent, it would have been obvious to one of ordinary skill in the art at the time the invention was made to add an anti-fading reagent to the prepared, stained slide in order to prevent the fluorescence from fading. One of ordinary skill in the art would have been motivated to use an anti-fading reagent in order to preserve the fluorescence of the stain so that it may be viewed long after mounting, as fluorchromes have a tendency to fade over a relatively short period of time. One would have expected success because anti-fading agents are commonly used in the art for this very purpose; for example, DakoCytomation sells a fluorescence mounting medium specially designed for cell specimens stained with fluorescent stains; it enhances visualization when viewed under a fluorescence microscope, and retards fading of the fluorescence (Claim 5). Therefore the invention as a whole would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made.

Claims 1-3 and 5 are rejected under 35 U.S.C. 103(a) as being unpatentable over Molecular Probes "MitoTracker and MitoFluor Mitochondrion-Selective Probes" Product Data Sheet and Molecular Probes "ER-Tracker Blue-White DPX" Product Data Sheet, in view of DakoCytomation.

Applicant's claim 1 is directed to a test preparation for microscopes comprising an object carrier and a biological cell bond arranged on the object carrier, wherein the cell bond is fixed with a compound which

enables a freely selectable fluorescence excitation in a broad range of wavelengths, including 100 nm and greater. The cell bond is being interpreted to mean a cell or tissue, comprised of cells. Claim 2 requires the fluorescence excitation range to be from 450 to 650 nm. Claim 3 requires the fluorescence excitation range to be from 350 to 700 nm. Claim 5 requires an anti-fading reagent to be added to the compound.

Molecular Probes teach preparations of cells stained with fluorescent probes; fluorescing cells (which applicant calls cell bonds) on slides read on what applicant calls test preparations for microscopes. Molecular Probes sell fluorescent probes that can be used to produce slides with stained cells for visualization of cellular components. For example, the family of MitoTracker Dyes are mitochondrion-selective probes that can be used to produce slides that show the mitochondria. Molecular Probes teach incubating cells in presence of Orange-, Red-, Deep Red-, and Green-Fluorescent MitoTracker Dyes and then fixing the cells in an aldehyde-based fixative (See Pg 1, col. 2- Pg. 2, col. 1). MitoTracker Orange exhibits fluorescent excitation at 554 nm; MitoTracker Red exhibits fluorescent excitation at 579 nm; MitoTracker Deep Red exhibits fluorescent excitation at 644 nm; and MitoTracker Green exhibits fluorescent excitation at 490 nm (See Table 1, Pg. 2) (Claims 1-3). Another example, ER-Tracker Blue-White DPX stains the endoplasmic reticulum of eukaryotic cells. Molecular Probes teaches adherent cells can be grown on a coverslip, which acts as an object carrier; incubated with the probe-containing medium for approximately 30 min (See Pg. 1, col. 2). The cells can then be fixed to the cover slip by application of 3.7% formaldehyde for 10-20 min (See Pg 2, col. 2). Fluorescence emission peaks anywhere in the range from 430-640 nm (See Pg. 1, col. 2) (Claims 1-3).

Though Molecular Probes does not teach use of an anti-fading reagent, it would have been obvious to one of ordinary skill in the art at the time the invention was made to add an anti-fading reagent to the prepared, stained slide in order to prevent the fluorescence from fading. One of ordinary skill in the art would have been motivated to use an anti-fading reagent in order to preserve the fluorescence of the stain so that it may be viewed long after mounting, as fluorchromes have a tendency to fade over a relatively short period of time. One would have expected success because anti-fading agents are commonly used in the art for this very purpose; for example, DakoCytomation sells a fluorescence mounting medium specially designed for cell specimens stained with fluorescent stains; it enhances visualization when viewed under a fluorescence

microscope, and retards fading of the fluorescence (Claim 5). Therefore the invention as a whole would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made.

Conclusion

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Allison M Ford whose telephone number is 571-272-2936. The examiner can normally be reached on M-F 7:30-5.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Michael Wityshyn can be reached on 571-272-0927. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306.

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Allison M Ford
Examiner
Art Unit 1651

LEONARD B. LANKFORD, JR.
PRIMARY EXAMINER